LA JOLLA CANCER RESE :H FOUNDATION

AUF 1413

ANIMAL USAGE FORM

TYPE OR PRINT

1. PRINCIPAL INVESTIGATOR	OFFICE PHONE	HOME/EMERGENCY PHONE
WAYNE A. BORDER, M.D.	226	(714) 770-4602
2. OTHER INVESTIGATOR		
LUCIA LANGUINO, PH.D.	230	539-0609
3. SENIOR TECHNICIAN		

4. PROJECT TITLE

ANTI-HUMAN TGFß CYCLIZED PEPTIDE

5. GRANT NUMBER, IF ANY	NEW	RENEWAL	PILOT	PROJECT NUMBER
250200	X			

6. START DATE	END DATE	QUANTITY: MICE	RATS	RABBITS	GPs	OTHER (SPECIFY)
				2		

7. PROJECT GOAL (SEE INSTRUCTIONS)

To produce quantities of anti-human TGFß cyclized peptide for use in kidney disease research.

8. RATIONALE (SEE INSTRUCTIONS)

Rabbits produce high quality antiserum which can be used for identification of human TGFß in tissue samples and in vitro assays to study progression of kidney injury.

9. DESCRIBE USE OF ANIMALS (SEE INSTRUCTIONS)

All injections/bleedings to be performed by animal care facility personnel.

1. Pre-bleeding 20 ml from ear vein .
2. Inject 500 µg TGFß cyclized purified peptide (0.5 ml antigen in PBS + 0.5 ml FCA) subcutaneously in 2 sites.
3. After one month, boost with 125 µg antigen (0.25 ml antigen in PBS + 0.25 ml incomple adjuvant) subcutaneously, 2 sites.
4. After 10 days, bleed 50 ml from alternating ear veins 3 times.
5. Repeat steps 3-4 at 4-6 week intervals.

RESEARCH PROTOCOL

10. PAIN LEVEL A [ ] B [ ] C [ ] (IF B OR C READ INSTRUCTIONS. PROVIDE DESCRIPTION OR JUSTIFICATION HERE)

CONFIDENTIAL

11. EUTHANASIA (SEE INSTRUCTIONS)

DURING PROJECT		METHOD OR TECHNIQUE	CO.		CERV. DISLOC.		RETAIN CARCASSES?	YES	
END OF PROJECT	X		O.D.	X	OTHER (SPECIFY)		FOR PI	NO	X

12. SIGNATURES

PI: WA Border DATE

U2 05334 DATE

AP MCA DATE

NIMAL PROCEDURE REQUEST

LJCRF

or procedures to be performed by Animal Facility personnel)
turn this form to the Animal Facility Office.

TODAY'S DATE: _____

Incipal Investigator E. RUOSLAHTI	Phone	Lab No.
LUCIA R LANGUINO	230	
vestigator/Technician	Phone	Lab No.

his procedure relates to the project outlined on Animal Usage Form No. _____

te or Dates on which this procedure is required (please allow 7 days for scheduling):

12/13/88	50 ml	(If more than 8 procedures are required, attach a second form)	5	
12/16/88	"		6	
12/21/88	Extra 100 ml		7	
			8	

Describe the procedure(s) required:

Vol. of blood to be collected each procedure: _____ ml.

Injection of 2 rabbits with Linear 78-103 peptide n°86-82 from TGFβ.

2 " with lyophilized 78-103 n°88-83

Procedure: 2 mg / rabbit of each peptide

- 2 mg peptide were dissolved in 250 µL DDW
- 0.5 mg Methylated BSA were added to the solution to couple the peptide (Sigma A1009 : MeBSA)
- vortex 30 "
- Added 250 µL Freud's adjuvant complete!
- Mixed 1 h with homogenizer.
- 0.5 ml solution was injected in each rabbit.

Note : Both peptides were difficult to dissolve.
" were not HPLC purified

Injection of 2 rabbits linear TGF β peptide

" Cyclic "

" PG peptide

Procedure

except Freund's incomplete adjuvant was used

For unique: KLH-peptide was already coupled
I only mixed to F.i.A. [illegible] 1h r.c
inject 0.5 ml/[illegible]

Rabbits n° 1284 for PG AIF 1:85
1285